Supplemental Figure legends

Supplemental Figure 1. Screening data pertaining to hybridomas.
First, 55 of 800 clones were selected for screening; subsequently, 24 of the 55 clones were selected owing to their viability and high reactivity with Gdn(-) or (+) materials. Among these 24 clones, 3 predominantly reacted with the Gdn(-) antigen. The characteristics of clone 1B12 changed during the limited dilution, probably as a result of isolation.

Supplemental Figure 2. Immunoblotting of PrPs with generated mAbs.
MAbs 2C4, 3B7, 3H6, and 4G4 (not reported in this paper because of instability of hybridoma) and 1B12 did not react with denatured PrPs. In contrast, mAbs Y3A6, X5C12, and X2H6, which were highly reactive to the Gdn(+) antigen, as determined using the screening tests (Suppl. Fig 1), reacted with PrPs in immunoblots like mAb T2.

Supplemental Figure 3. Epitope mapping by peptide array.
Reactivity of generated mAbs to synthetic peptide arrays on the cellulose support used for epitope mapping in a previous study (1) were shown. In the case of the generated mAbs, no positive spot was observed after long exposure to the arrays. In contrast, several spots were detected in the case of the non-conformational mAb X2H6 (Suppl. Figs. 1 and 2) after a short exposure.

Supplemental Figure 4. PrPs immunoprecipitated from the brain homogenates of various species.
Representative immunoblots used to calculate the data shown in Table 2.

Supplemental Figure 5. PK resistance of PrP^{core} analyzed using immunoblotting.
Immunoblotting was performed using PrP^{core} samples that were obtained from serially passaged mice and subjected to long-term PK (LTPK) digestion as described in Experimental Procedures. *A*. Immunoblots of PrP^{core} subjected to LTPK digestion. *B*. Degradation of PrP^{core} after LTPK digestion. The average intensity of each band calculated from the results of two separate experiments has been plotted. Degradation curves were classified into two groups based on the passage number (second and third passages) as well as the results of ELISA (Fig. 2).

Supplemental Figure 6. Sequential immunoprecipitation of PrP^{Sc} obtained from mice in the 3rd passage.
The PrP^{Sc} in the unbound supernatant after the first immunoprecipitaion assay was re-precipitated with an equal amount of mAb 3H6 or concentrated using a 2-butanol/methanol mixture (2) and detected by immunoblotting. The amount of PrP^{Sc} bound to mAb 3H6 decreased even though a sufficient quantity of PrP^{Sc} was present in the supernatant.

Supplemental Figure reference

1. Yokoyama, T., Kimura, K. M., Ushiki, Y., Yamada, S., Morooka, A., Nakashiba, T., Sassa, T., and Itohara, S. (2001) *J. Biol. Chem.* **276**, 11265–11271

2. Iwata, N., Sato, Y., Higuchi, Y., Nohtomi, K., Nagata, N., Hasegawa, H., Tobiume, M., Nakamura, Y., Hagiwara, K., Furuoka, H., Horiuchi, M., Yamakawa, Y. and Sata, T. (2006) *Jpn. J. Infect. Dis*. **59**, 100-107

Supplemental Figure1.

	clone		OD450	
			Gdn(+)	Gdn(-)
1	X	1E12	3.225	0.128
2	X	1B12	1.232	0.390
3	X	2H6	3.297	3.200
4	X	2G7	3.537	0.643
5	X	3H9	3.401	0.276
6	X	4H5	0.134	0.120
7	X	4E9	0.765	0.787
8	X	4F9	0.117	1.433
9	Y	5B4	0.127	0.118
10	X	2C4	0.335	1.933
11	X	3B1	0.559	0.330
12	X	3B7	0.127	1.130
13	X	4G4	0.483	1.269
14	X	4A8	3.431	1.473
15	X	4B8	3.416	1.501
16	Y	2C5	0.978	0.519
17	Y	2F5	3.267	3.280
18	Y	2F6	1.006	1.066
19	Y	2H10	3.450	3.334
20	Y	3D10	3.472	3.190
21	Y	4H10	3.331	0.383
22	X	2H2	3.357	3.318
23	X	3H6	0.433	2.782
24	X	4F7	3.362	3.152
25	X	5C12	3.316	3.662
26	Y	1E8	3.215	3.414
27	Y	1B12	3.554	3.290
28	Y	2F9	0.340	1.017
29	Y	3F1	3.554	3.214
30	Y	3A6	3.538	3.394
31	Y	5F4	3.399	3.308
32	Y	5G4	3.166	3.296
33	X	1H1	0.094	0.091
34	X	1F3	0.111	0.097
35	X	1H4	0.120	0.109
36	X	1G9	0.127	0.119
37	X	1G12	1.695	1.080
38	X	2E9	1.640	0.132
39	X	2F9	2.654	1.387
40	X	2G9	0.232	0.148
41	X	3G4	3.023	2.476
42	X	3H5	3.443	2.323
43	X	3D12	3.229	3.231
44	X	4C1	0.387	0.361
45	X	4H10	3.085	2.331
46	X	5A11	0.361	0.257
47	X	5A12	0.096	0.101
48	Y	5G6	1.108	0.809
49	Y	1B4	3.346	3.018
50	Y	2A6	2.882	2.337
51	Y	2F11	3.373	3.388
52	Y	3G2	3.284	3.332
53	Y	3E5	3.245	3.285
54	Y	3F10	3.297	3.405
55	Y	4D12	3.348	3.330

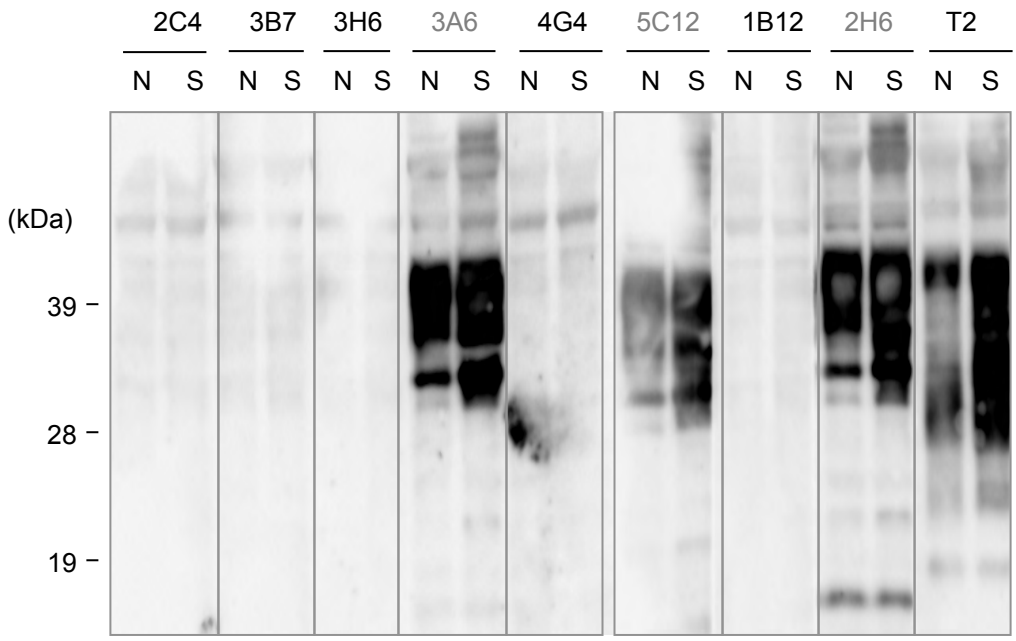
	clone		OD450	
			Gdn(+)	Gdn(-)
1	Y	1B12*	1.064	2.143
2	Y	1E8	3.215	3.414
3	X	1G12	1.695	1.080
4	X	2C4	0.335	1.933
5	Y	2F11	3.373	3.388
6	Y	2F6	1.006	1.066
7	Y	2F9	2.654	1.387
8	Y	2H10	3.450	3.334
9	X	2H6-1	3.297	3.200
10	Y	3A6	3.538	3.394
11	X	3B7	0.127	1.130
12	Y	3D10	3.472	3.190
13	X	3D12	3.229	3.231
14	Y	3F1	3.554	3.214
15	Y	3F10	3.297	3.405
16	Y	3G2	3.284	3.332
17	X	3H5	3.443	2.323
18	X	3H6-1	0.433	2.782
19	Y	4D12	3.348	3.330
20	X	4F7	3.362	3.152
21	X	4F9-2**	0.117	1.433
22	X	4G4-1**	0.483	1.269
23	X	5C12	3.316	3.662
24	Y	5G4	3.166	3.296

Gdn(+)<Gdn(-)

*data of after 4th cloning

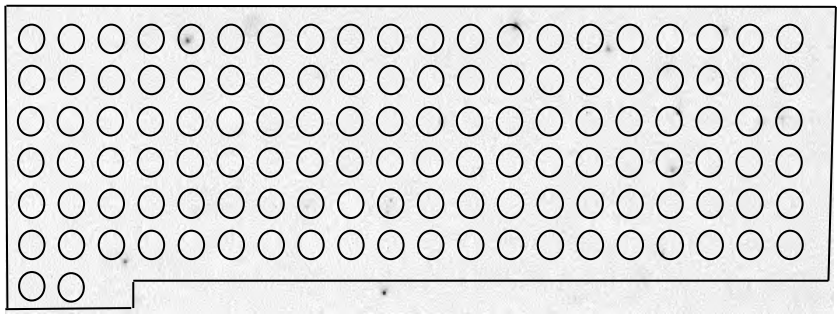
** unstable clones

Supplemental Figure 2.

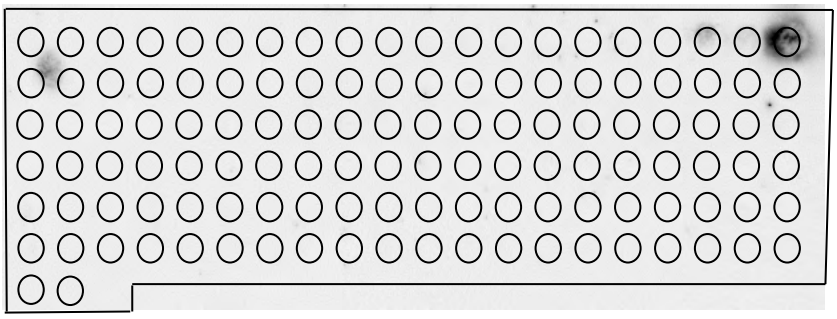


Supplemental Figure 3.

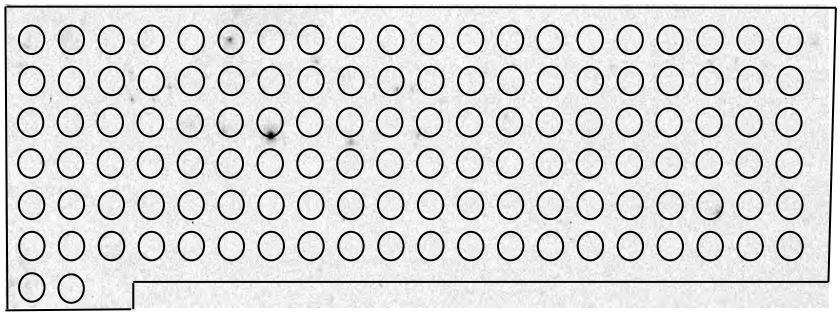
2C4



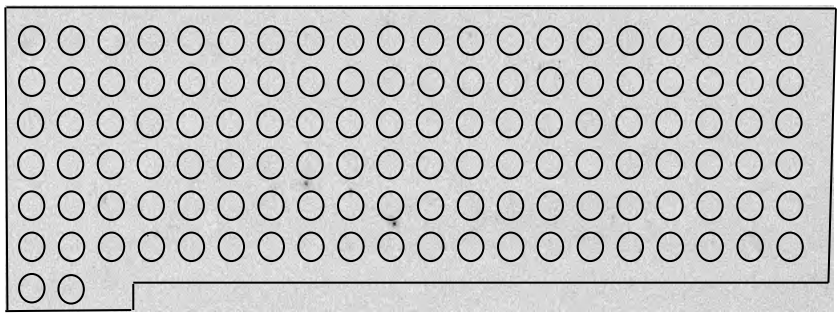
3B7



3H6

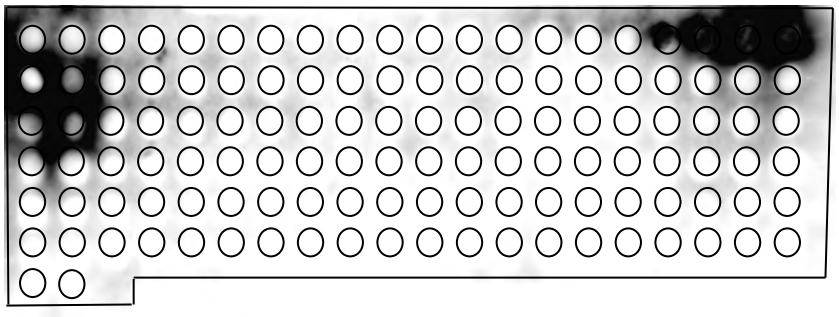


1B12



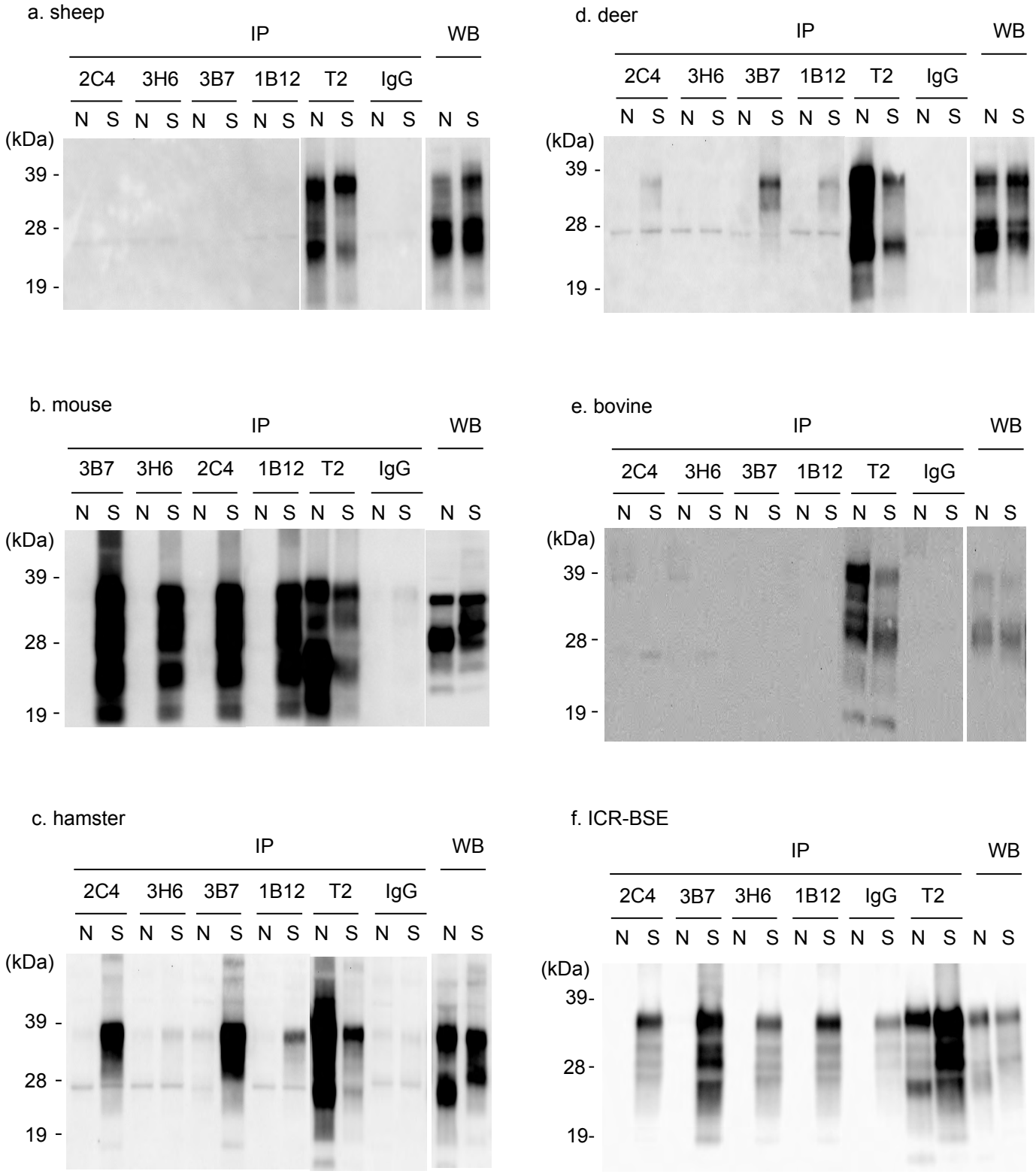
Long exposure: 10min.

2H6



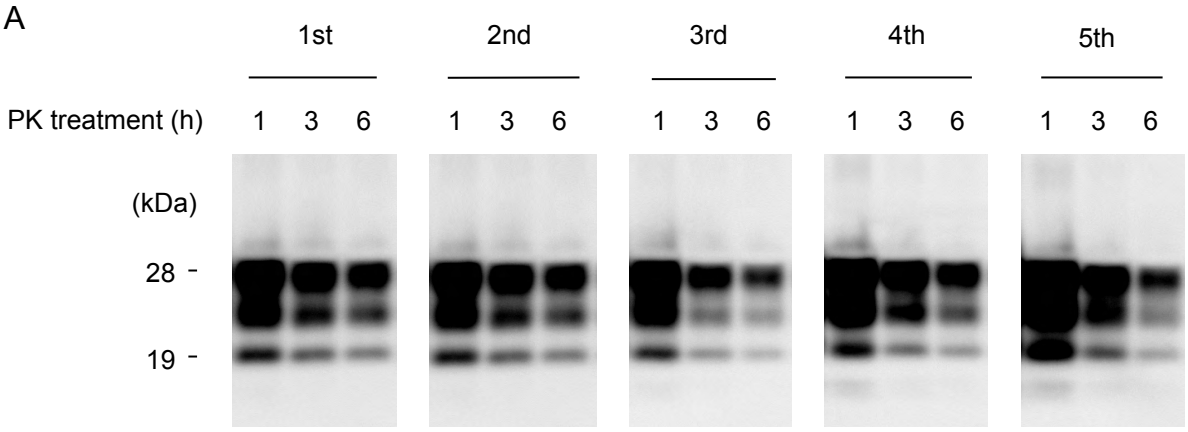
Exposure time: 30sec.

Supplemental Figure 4.



Supplemental Figure 5.

A



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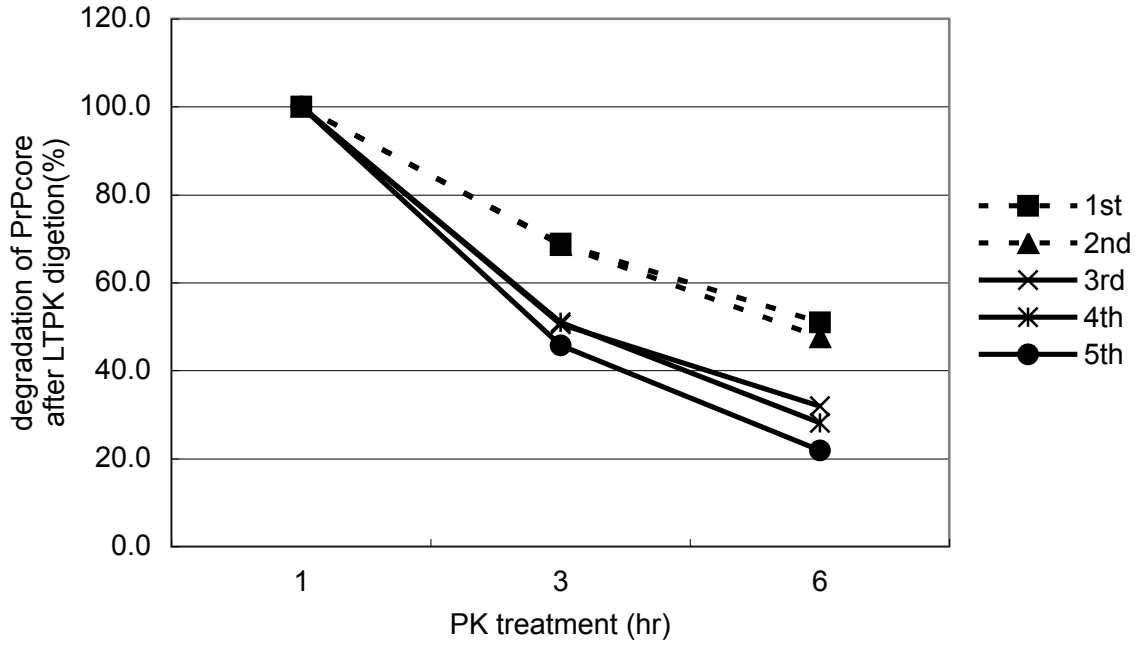

Supplemental Figure 6.